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Role of Kombucha Tea in the Control of EMF 950 MHz Induced Injury in Rat Heart and Lung Organs

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Key Words

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Abstract

Objective: Cell phone usage has increased significantly nowadays. The present study has highlighted the negative effect of cell phone exposure in rats and the possible protective role of Kombucha tea to overcome these effects.

Methods: The control group received a black tea only, while KT group received KT (0.1ml/100g BW/day) orally, the EMF group exposed to EMF (950MHz) 1 hr daily for 50 days, the KT- EMF group were pre- treated with KT for one week and then simultaneously challenged with EMF exposure for additional 50 days. Cardiopulmonary tissue oxidant/antioxidant status was determined.

Results: EMF exposure caused marked rise in cardiopulmonary lipid peroxidation (MDA), superoxide dismutase (SOD) besides serum lactate dehydrogenase (LDH) and aspartate amino transferase (AST) levels whereas tissues glutathione peroxidase (GSH-Px) as well as serum total antioxidant capacity were decreased. KT administration successfully attenuated these adverse effects of EMF.

Conclusions: Our study demonstrated that KT could proffer a measure protection against EMF exposure induced oxidative damage by possibly reducing lipid peroxidation and increasing the antioxidant defense mechanism in rats.

INTRODUCTION

In available literature there are data suggesting that exposed to strong electromagnetic fields can produce an increased amount of reactive oxygen species in tissues, resulting in stimulation of peroxidation of membrane lipids leading to apoptosis and death of cells^[1,2]. Recent attention has focused on non-ionizing, non-thermal electromagnetic fields as an environmental etiology of human disease. Research suggests that electromagnetic fields may influence the immune system^[3], behavior^[4], pregnancy^[5], chronobiology^[6], and cancer^[7]. Magnetic fields have been found to affect significantly the cardiac function; in addition not all magnetic fields are the same. Different types of magnetic fields may have different effects on the heart. Participation of free radical biochemistry in several pathologies and aging has been demonstrated. Free radicals, such as superoxide anions (O⁻ 2), generated by electrical stimuli show high chemical reactivity and, as a result, have a relatively short lifetime in the free state^[8]. Free radical oxidation of polyunsaturated fatty acids in

biological system is known as lipid peroxidation, and detection and measurement of lipid peroxidation is the evidence most frequently cited to support the involvement of free- radical reactions^[9]. Free radicals are very reactive and unstable molecular species that can initiate chain reactions to form new free radicals. Several mechanisms are in place to neutralize their effects, which include a system of nutritional and endogenous enzymatic antioxidant defenses that generally hold the production of free radicals and prevent oxidant stress and subsequently tissue damage^[10,11]. SOD scavenges the superoxide radicals by catalyzing the reactive O⁻ 2 species into dioxygen and oxygen peroxide, thereby protecting cells against the reactive oxygen species (ROS) produced by electric fields or other mechanisms^[12-18]. Kombucha tea (KT) is a sugared black tea fermented with tea fungus for about 10 days. Tea fungus is a symbiotic culture of acetic acid bacteria and yeasts. KT is claimed to have various beneficial effects on human health, but there is very little scientific evidence available in the literature. Kombucha tea is composed of two portions; a floating cellulosic pellicle layer and the

under lying sour liquid broth, which tastes slightly sweet and acidic^[19]. Recent studies have suggested that kombucha tea prevents paracetamol-induced hepatotoxicity^[20] and chromate (VI) and radiation-induced oxidative stress in albino rats^[21, 22]. As kombucha tea is rich in compounds known to be strong antioxidants, it is expected to ameliorate cardiopulmonary damage induced by EMF.

Hence, in the present study, the prophylactic and curative effect of kombucha tea is tested against EMF induced cardiopulmonary damage using male albino rats as an experimental model.

MATERIALS AND METHODS

Animals

Male Wister rats each weighing 150-180 gm were purchased from the animal breeding unit of National Center of Radiation Research and Technology they were housed under appropriate conditions of constant humidity temperature of $22^{\circ}\pm 2^{\circ}$ and light. The animals were allowed free access to water; they were fed a standard pellet rat diet. The rats were housed for at least 1 week before subjecting them to experimentation. The study was approved by the Ethical Committee for Animal Experimentation at the National Center for Radiation Research and Technology.

Preparation of Kombucha Tea

The tea fungus was obtained from our microbiology lab of the National Center for Radiation Research and Technology (NCRRT), Atomic Energy Authority (Cairo, Egypt) where the author is employed. It was grown and maintained in tea medium^[21].

Experimental Design

In the control group, the animals were administered with black tea with the stomach tube throughout the experimental period. KT administered group, the animals were administered with Kombucha ferment tea (0.1ml/ 100g of b.w. /day) orally by means of stomach tube for 57 days. In the exposed group, animals were exposed to 950 MHz for 1h daily for 50 days together with black tea administration orally. In KT& EMF treated group, the animals' administered with KT for one week, then the animals exposed to the EMF (950MHz) concurrently with KT administration for another 50 days. Control and KT groups were kept

under the same conditions without being exposed to the electromagnetic wave. At the end of the experiment, the experimental and control animals were weighed and anesthetized with ether. The blood sample was collected from left ventricle of the heart. Heart and lung were quickly separated and cleaned in normal saline solution then weighted individually for biochemical analysis.

Microwave System

The exposure device was designed and constructed locally in the Radiation Physics Department; NCRRT, Cairo, Egypt; to study the biological effect between 0.01 and 20 GHz. The microwave exposure system consisted of the following main parts (Figure. 1). The microwave generator (HP 83712B) with frequencies range between 0.01 and 20.0 GHz. HP 8592L Spectrum analyzer that cover range from 9 KHz to 22 GHz. Two horns antennas, one working as a transmitter and the other as a receiver.

All animals were housed collectively (8 animals in each wooden cages $30 \times 40 \times 40$ cm- W \times L \times H). The animals were exposed to microwave 950 MHz at specific absorption rate (SAR) for 50 days (1 hr daily during the light period). SAR was calculated as the following equation:

$$\text{Power density (Pd)} = \frac{GP}{4\pi R^2}$$

Where, G= Gain of the antenna.

P= Power of the antenna.

R= Distance between antenna and ample. At frequency 950 MHz, the Pd is equivalent to 1W/Kg of SAR.

Biochemical Analysis

All biochemical assays were performed with a Helios Thermo-Spectronic spectrophotometer (Thermo Spectronic, UK). Total antioxidant capacity was analyzed by Koracevic et al.^[23]. Lactate dehydrogenase (LDH) activity was evaluated according to the method recorded by IFCC^[24]. AST activity was evaluated according to the method of Reitman and Frankel^[25] with a Bio-Diagnostickit. Total protein of heart and lung was measured according to the method by Lowery et al.^[26]. Concentration of heart and lung malondialdehyde (MDA) was analyzed according to the method recorded by Yoshioka et al.^[27]. Both heart and lung homogenate SOD activities were measured according to the method by Marklund and marklund^[28]. Glutathione peroxidase activity was measured according to the method by

Lawrence and Burk ^[29]. Catalase activity was measured according to the method of Sinaha ^[30].

Statistical Analysis

Quantitative data were expressed as mean ± standard error (SEM) and were analyzed by one way analysis of variances (ANOVA) followed by Tukey's multiple comparison test. Data are representative of 8 independent experiments carried out in triplicate. Statistical analysis was performed by using Graph-Pad software, San Diago, CA, USA) Differences were considered statistically significant when the P value was less than 0.05.

RESULTS

As reported in previous studies, EMF

exposure induced structural alteration in both heart and lung organs resulting in a significant reduction in total antioxidant capacity (P= 0.001), cardiopulmonary GSH-Px activity (P= 0.01, 0.001 respectively) as shown in Figure. 2&3 accompanied by significant increases in cardiopulmonary MDA concentration (P= 0.001) and serum LDH (P= 0.001) and AST (P= 0.001) (Figure. 4). In contrast, in EMF exposed animals, KT administration was associated with a significant increase in total antioxidant capacity, cardiopulmonary GSH-Px activity (Figure. 4&3). Likewise, KT significantly reduced the serum LDH and AST as well as cardiopulmonary SOD activities besides cardiopulmonary MDA concentration bringing back to levels similar to un-irradiated control animals besides (Figure. 4).

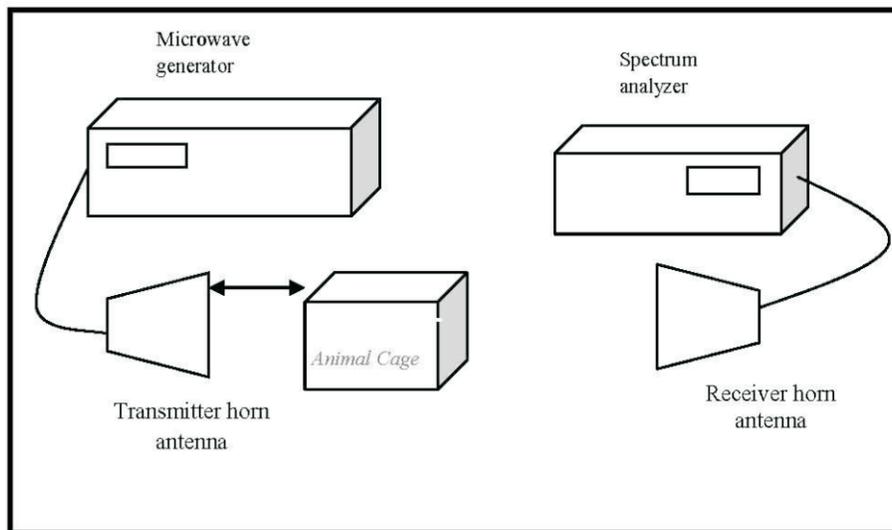
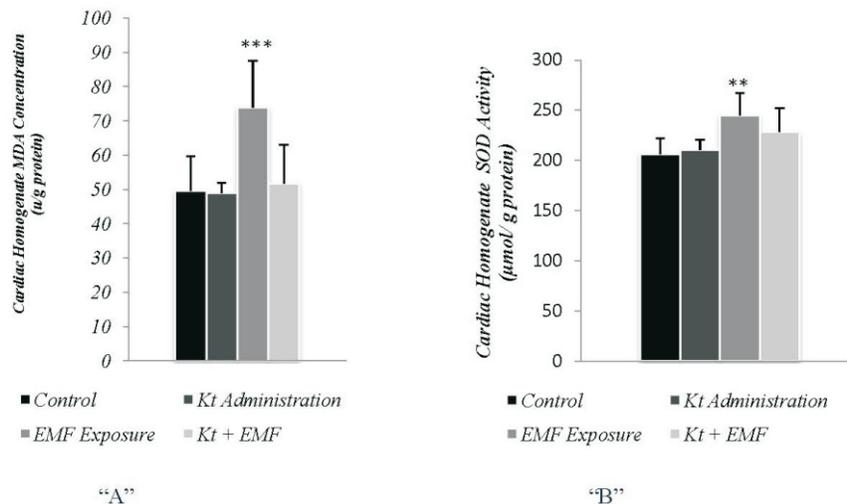


Figure 1: Schematic Diagram of EMF Instruments



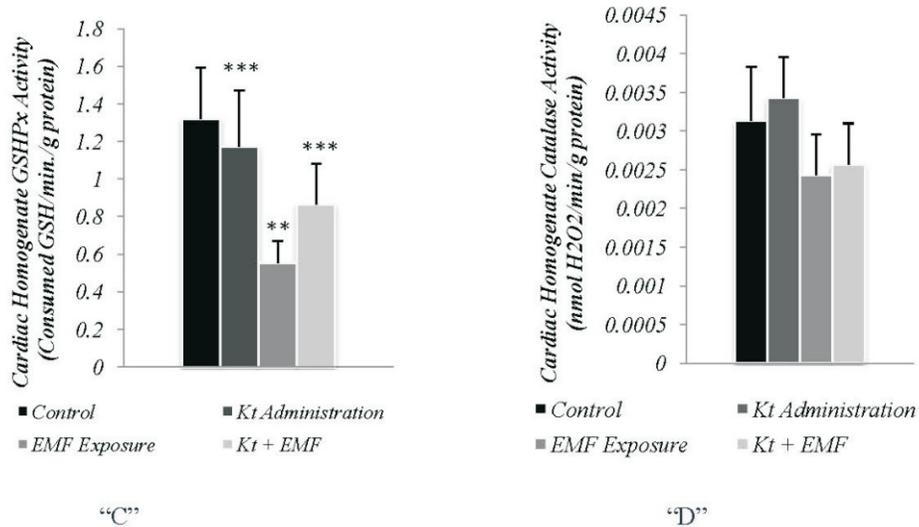


Figure. (2): Effect of Kombucha tea pre-treatment (0.1ml/ 100g b.w. 50 days) on cardiac homogenate of MDA concentration ($\mu\text{mol/g protein}$) (A), SOD activity ($\mu\text{mol/g protein}$) (B), GSH-px activity (consumed GSH/min./g protein) (C) and Catalase activity (nmol H₂O₂/min./g protein) (D).

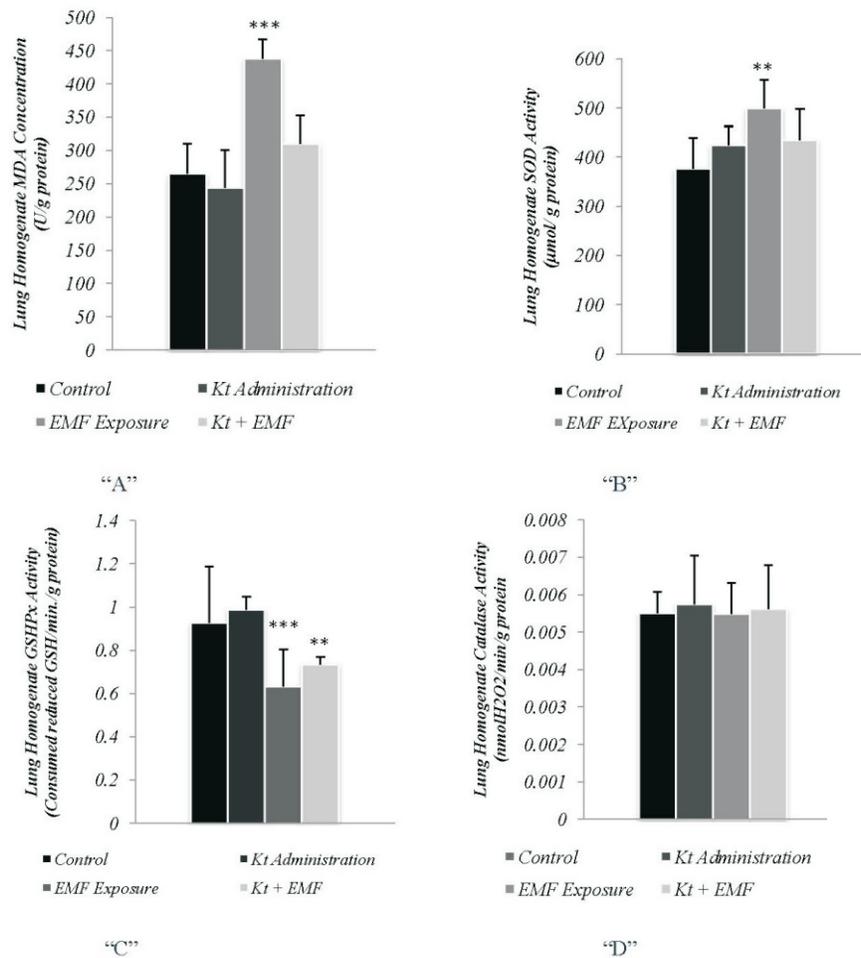


Figure. (3): Effect of Kombucha tea pre-treatment (0.1ml/ 100g b.w. 50 days) on lung homogenate of MDA concentration ($\mu\text{mol/g protein}$) (A), SOD activity ($\mu\text{mol/g protein}$) (B), GSH-px activity (consumed GSH/min./g protein) (C) and Catalase activity (nmol H₂O₂/min./g protein) (D).

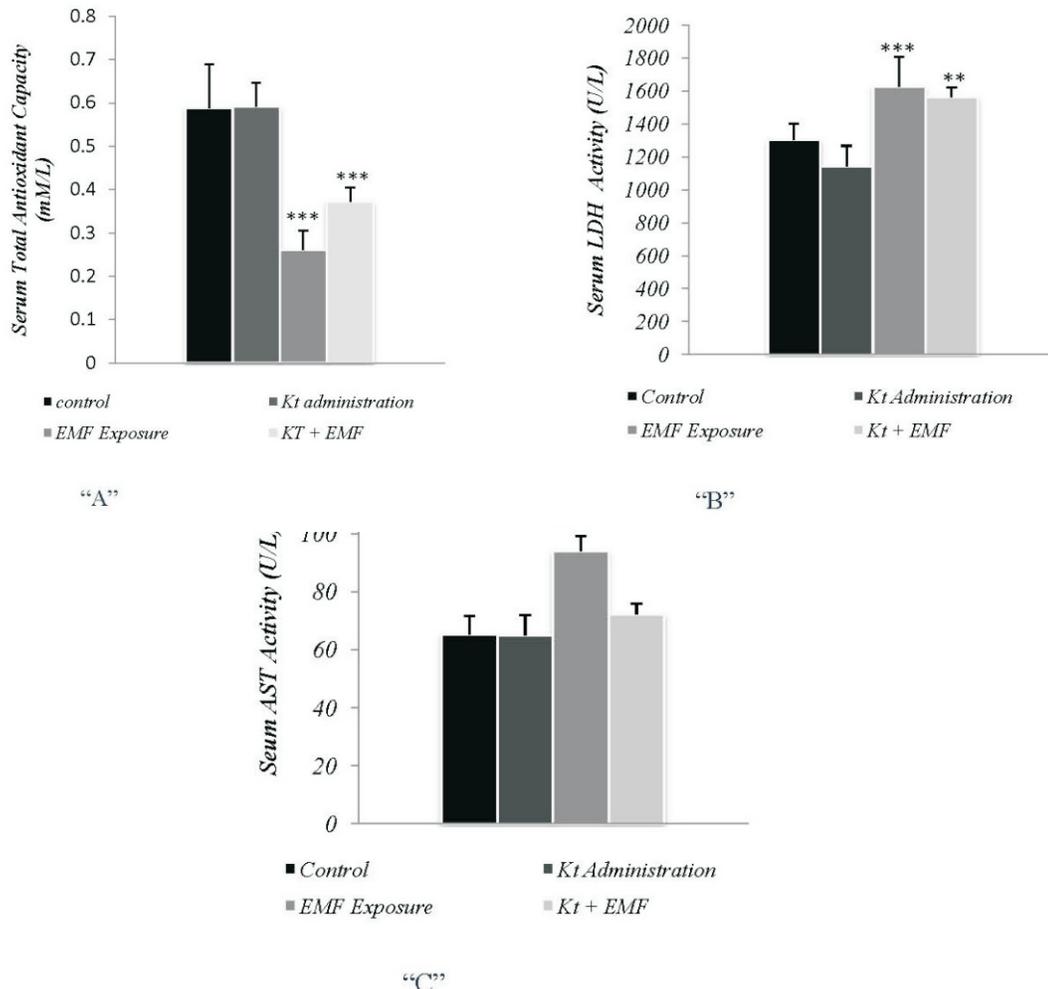


Figure. (4): Effect of Kombucha tea pre-treatment (0.1ml/ 100g b.w. 50 days) on serum total antioxidant capacity (mM/L) (A), LDH activity (U/L) (B) and AST activity (U/L) (C) ©.

DISCUSSION

Exposure to electromagnetic field (EMF) can lead to cell death as a result of the increase in free radicals and DNA damage^[8]. One hypothesis, based on well-established observation, is that magnetic field can increase the yields of some types of free oxygen radicals such as $O_2^{\cdot-}$ and OH^{\cdot} ^[15,31]. It has been demonstrated in numerous studies that ROS are directly involved in oxidative damage of cellular macromolecules such as lipid, proteins and nucleic acids in tissues^[18]. The main ROS that have to be considered are $O_2^{\cdot-}$ which predominantly generated by the mitochondria producing hydrogen peroxide (H_2O_2) by the action of SOD^[14]. ROS are scavenged by SOD, Glutathione Peroxidase (GSH-Px) and Catalase. Catalase, glutathione peroxidase and superoxide dismutase are antioxidant enzymes

which collaborating with each other protect the organism against toxic action of reactive oxygen species^[32,33]. Laboratory studies show that high frequency EMF affects the cardiovascular system. The cell membrane in primary site of EMF interaction, is leading to intercellular changes in genes function and protein synthesis^[34]. In this study, exposure to 950 MHz led to an increase in both serum LDH and AST activities and reduction in total antioxidant capacity due to the presence of free radicals involved in EMF-induced tissue injury^[10,14], accompanied by increase in both cardiopulmonary SOD and MDA concentrations, which is in agreement with Benov et al,^[12] & Yeh et al.^[35] who reported that MDA levels increase with the increase in SOD activity. Güler et al.^[16] hypothesized that the increase in MDA resulting from energy transfer in the tissue, might be due to

the transformation of molecular O₂ to free radical, thus increasing the level of O₂·. On the other hand, the increase in SOD activity might be regarded as an indicator of increased ROS production occurring during the exposure period and may reflect the patho-physiological process of the exposure. In the present study, exposure of animals to 950 MHz showed a significant reduction in GSH-Px activity, while Catalase activity level was not statistically significant which are free radical scavenger. These results are in agreement with the results obtained from Moustafa et al.^[11]. As reported by Gharib^[22] KT supplied the animals a protection against oxidative stress induced by radiation exposure. In the present study, administration of KT ferment concurrently with EMF exposure resulted in amelioration of the above monitored parameters. This effect might be due to the presence of B vitamins in KT ferment since they act as a protective agent against heart damage^[36]. Moreover, β-glucan, which is a by-product of the KT ferment, is a cell wall component, which is completely orally safe, potent free radical scavenger and non-specific stimulator of immune response^[37]. It also plays a pivotal role in the initiation and maintenance of the immune response. The presence of hyaluronic acids, which enable to lessen free radical damage; amino acids which are constituent of proteins, produce important enzymes, such as glutathione a powerful antioxidant which provides protection from the oxidative stress^[38] and the nucleic acids like RNA and DNA, that transmit information to the cells on how to perform correctly and regenerate^[39].

CONCLUSION

In summary, the present results suggest that KT ferment could act as a protective agent against the injury induced by EMF exposure, which is mainly induced by using mobile phones.

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